

## REMARKS

Applicant intends this response to be a complete response to the Examiner's **25 May 2010** Non-Final Office Action. Applicant has labeled the paragraphs in his response to correspond to the paragraph labeling in the Office Action for the convenience of the Examiner.

## DETAILED ACTION

The Examiner states and/or contends as follows:

1. The amendment, response and remarks filed 13 August 2010 to the Office Action with Non-Final Rejection mailed 25 May 2010 is acknowledged and entered.

Applicants acknowledge this statement.

### *References Cited in 371 Application*

The Examiner states and/or contends as follows:

2. In response to the request by the Examiner to list references/ "Documents submitted with 371 Application" Applicants have complied with 37 C.F.R. §1.98(a) (1) and provided copies of said references so that said references be printed on a resulting patent. Said references are listed on appropriate PTO form(s).

Applicants acknowledge the Examiner's statements and thank the Examiner.

### *Informals*

The Examiner states and/or contends as follows:

3. This application contains Claims 1-13 drawn to an invention nonelected with traverse in the reply filed on 25 February 2010. A complete reply to the final rejection must include cancellation of nonelected claims or other appropriate action.

Applicants have canceled the claims directed to a non-elected restriction, but expressly reserve the right to file divisional applications directed at the non-elected subject matter and any other subject matter not covered by the elected claims set.

## Claims Status

The Examiner states and/or contends as follows:

4. According to the amendment filed 13 August 2010, following is the status of the Claims:  
Claims 1-30 are pending;  
Claims 14,27 and 29-30 are currently amended;  
Claims 1-13 are withdrawn; and  
Claims 14-30 are currently under examination.

Applicants acknowledge these statements.

### ***Withdrawals***

The Examiner states and/or contends as follows:

5. Considering the Amendments and remarks filed 13 August 2010, the following objections and rejections in the Office Action mailed 25 May 2010 are hereby withdrawn:

- Objection to specification;
- Indefinite rejection of Claim 30 under 35 U.S.C. § 112, 2nd paragraph;
- Anticipatory, or in the alternative obviousness rejection of Claims 14-26 under 35 U.S.C. §§ 102(b)/103(a) by Logan (U.S. Patent 6,214, 607 B1) ;and
- Obviousness rejection of Claims 27-30 under under35 U.S.C. §103(a) over the combined teachings from Logan (U.S. Patent 6,214,607 B1) in view of Venkatesh et al (U.S. Patent 6,066,257).

Applicants acknowledge the withdrawals set forth above.

### ***Information Disclosure Statement***

The Examiner states and/or contends as follows:

6. The Information Disclosure Statements (i.e., IDSs) filed respectively on 21 June and 13 August 2010 have been made of record, considered and duly initialed sheets of the *PTO/SB/08A* are enclosed with this Office Action.

Applicants acknowledge the Examiner's statement.

### ***Claim Rejections - 35 U.S.C. §103***

The Examiner states and/or contends as follows:

7. Considering the amendment to Claims filed 13 August 2010, and references provided with the Information Disclosure statements filed respectively on 21 June and 13 August 2010; following are new art rejections to Claims 14-30.

Applicants acknowledge the Examiner's statement.

10. **Claims 14-26** stand rejected under 35 U.S.C. §103(a) as obvious over combined teachings of Logan (U.S. Patent 6,214,607 B1) in view of Okeke et al (2002. Reduction of perchlorate and nitrate by salt tolerant bacteria. ENVIRONMENTAL POLLUTION, Volume 118, Pages 357-363., Applicants' IDS filed 08/13/2010, Item 11).

The Examiner states and/or contends as follows:

Claims 14-26 are drawn to a method to degrade at least one contaminant present in a contaminated brine stream having "a salinity of  $\geq$  about 3%" when said stream is:

- fed to a bioreactor containing a mixed bacterial culture, wherein;
- said bacterial culture degrades under anoxic/anaerobic conditions said at least one pollutant in said brine stream;
- to said bioreactor is also added an effective quantity of a divalent cation precursor in such amount that said divalent cation quantity maintains a divalent to monovalent cation molar ratio at  $\geq 0.05$ ;

- said degradation of said brine stream contaminant takes place at a temperature for a duration to bring the contaminant concentration at a desired level, while divalent to monovalent cation molar ratio is maintained at  $\geq 0.05$ ;
- said bioreactor is sealed to eliminate/reduce oxygen;
- said reactor being sparged with an oxygen-free gas selected from nitrogen (N<sub>2</sub>) or Argon (Ar) or mixtures thereof;
- divalent cation selected from soluble salt of:
  - Ba<sup>2+</sup>, Ca<sup>2+</sup>, Mg<sup>2+</sup>, Sr<sup>2+</sup>; or
  - Ca<sup>2+</sup>, Mg<sup>2+</sup>, Sr<sup>2+</sup>; or
  - Ca<sup>2+</sup> and Mg<sup>2+</sup>; or
  - is Mg<sup>2+</sup>;
- the contaminant is selected from the group perchlorate (ClO<sub>4</sub><sup>1-</sup>) or nitrate (NO<sub>3</sub><sup>1-</sup>) or mixture thereof;
- nutrients comprising an inorganic, or organic nutrient source in an amount greater than a stoichiometric amount of electron required to reduce the ClO<sub>4</sub><sup>1-</sup> and/or NO<sub>3</sub><sup>1-</sup> contaminant present in the brine solution for sustained microbial growth;
- the inorganic energy source is selected from the group H<sub>2</sub> gas, a H<sub>2</sub> delivery chemical or mixtures thereof;
- the organic nutrients are selected from the group consisting of acetate, ethanol, lactate, methanol, or mixtures thereof; and
- the contaminated brine solution is a ClO<sub>4</sub><sup>1-</sup> and/or NO<sub>3</sub><sup>1-</sup> contaminated ion-exchange regenerate brine.

Regarding Claims 14-26, Logan teaches degradation of ClO<sub>4</sub><sup>1-</sup> in a brine solution, comprising per liter of deionized water: 5.85 mg of NaCl, 6.5 mg of MgSO<sub>4</sub>, 4.3 mg of CaSO<sub>4</sub>•2H<sub>2</sub>O and 48.0 mg of CaCO<sub>3</sub> with 100 mg acetate, nitrogen and phosphorus (Example 1; Column 8, Lines 40-45). Logan further teaches that degradation studies for said ClO<sub>4</sub><sup>1-</sup> contained in said brine solution was carried out in crimp-topped serum vials (i.e., bioreactors), whose head space was purged with N<sub>2</sub> gas, said bioreactors inoculated with ClO<sub>4</sub><sup>1-</sup> degrading mixed microbial consortium and the degradation was carried out for 7 days at room temperature. In 4 days, 39% ClO<sub>4</sub><sup>1-</sup> was removed (Example 1; Column 8, Lines 25-40). Subsequently column reactors were inoculated with the same microbial consortium wherein the column reactors contained the same ClO<sub>4</sub><sup>1-</sup> containing brine solution described *supra* (Example 1; Column 8, Lines 40-45). Logan further teaches that the pH of the column was in range of 5.0-8.0, the column temperature 10°C to 30°C to keep bacterial cells viable and active, and the system is comprised of an "oxidizable substrate serving as electron donor." Oxidizable substrates include acetate, ethanol, methanol and hydrogen gas (Column 6, Lines 8-20). Based on the concentrations of each of NaCl, MgSO<sub>4</sub>, CaSO<sub>4</sub>•2H<sub>2</sub>O and CaCO<sub>3</sub> said brine solution has a divalent to monovalent cation ratio of 0.1060. Thus, Lawrence et al., teach each and every limitation: H<sub>2</sub> gas as an inorganic nutrient, acetate, methanol, or ethanol as an organic nutrient, purging with N<sub>2</sub> gas, ClO<sub>4</sub><sup>1-</sup>-contaminated brine solution having divalent to monovalent cation ratio of 0.1060 (i.e.,  $\geq 0.05$ ), and 39% degradation of ClO<sub>4</sub><sup>1-</sup> of the invention claimed in Claims 14-26.

Logan, however, is silent regarding the word brine, wherein said brine stream has "a salinity of  $\geq$  about 3%" as required in instantly amended Claim 14.

Okeke et al., teach perchlorate and nitrate removal/degradation in a bacterial culture medium comprising: K<sub>2</sub>HPO<sub>4</sub>, KH<sub>2</sub>PO<sub>4</sub>, (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, MgSO<sub>4</sub>•7H<sub>2</sub>O, CaCO<sub>3</sub>, FeCl<sub>3</sub>•4H<sub>2</sub>O and acetate (Page 358, Column 2, Lines 5-11). Okeke et al., further teach that in a comparative study between *Citrobacter* sp. and perclace (ATCC 202172), effect of sodium chloride at concentrations of 0%, 2.5%, 5% and 10% was examined (Page 359, Column 1, Lines 39-43, section 2.4.3). Okeke et al., also teach a contaminant brine stream having 100 mg/mL perchlorate in final concentration and further inoculated with co-culture comprising *Citrobacter* sp. and perclace (Page 359, Column 2, Lines 5-11).

It would have been *prima facie* obvious to a person of ordinary skill in the art and one having ordinary skill in the art at the time of the claimed invention would have been motivated to modify/combine the teachings from Logan with those of Okeke et al., to obtain a method wherein at least one contaminant present in a contaminated brine stream having "a salinity of  $\geq$  about 3%" when

said stream is fed to a bioreactor containing a mixed bacterial culture, is decontaminated or at least reduced in level of at least one contaminant (*i.e.*, perchlorate); because Okeke at al., teach a contaminant brine stream having 100 mg/mL perchlorate in final concentration and said stream further comprises a co-culture comprising *Citrobacter* sp. and Perclace, wherein said co-culture reduces the contaminant/perchlorate in said stream. The recited prior art may be silent regarding the exact technique as are instantly claimed, however, since similar steps and same components and principle are described in the prior art references, application of those parameters and steps is deemed merely a matter of judicious design selection of a result-effective parameter which is well within the purview of the skilled artisan especially with the relevant art from the cited references before him/her as a guide (See, e.g., M.P.E.P. §2144.06) and is therefore obvious under 35 U.S.C. §103(a).

From the teachings of the cited references, it is apparent that one of ordinary skill in the art would have had a reasonable expectation of success in producing the claimed invention. Therefore, the invention as a whole was *prima facie* obvious to one of ordinary skill in the art at the time the invention was made, as evidenced by the references, especially in the absence of evidence to the contrary.

Applicants do not understand the Examiner's reading of Logan. Logan is not directed to brine. The Examiner suggests that Logan does not include the word brine as if it were an oversight, but Logan is direct to treating ground water to used for drinking to reduce perchlorate concentrations. Thus, Logan never discussed brines because Logan is directly specifically and expressly to drinking water. In fact, the water that the Examiner relies on to support a teaching of the divalent to monovalent metal ratio is an artificial ground water based on the composition of granitic rock aquifer, a source of drinking water:

**An artificial groundwater (AGW) based on the composition of granitic rock aquifer was used in column studies.** The water consisted per liter of deionized water, of: 6.5 mg of  $\text{MgSO}_4$ , 4.3 mg  $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$ , 5.85 mg of NaCl, and 48.0 mg of  $\text{CaCO}_3$ . To this AGW, the following was added in column biodegradation studies: 100 mg/L acetate, 20 mg/L perchlorate, and nitrogen and phosphorus at a ratio of 100:23:4.3 as C:N:P. All samples were immediately filtered through 0.2 mm polycarbonate filters (Poretics Corp.) to remove bacteria prior to chemical analysis.

Logan at Col. 8, ll. 39-48 (emphasis added).

Moreover, Logan does not disclose, teach or even lead an ordinary artisan to expect that by producing a specific ratio of divalent to monovalent metal cations in a brine would produce a solution capable of supporting a stable microbial population, where the stable population is capable of reducing perchlorate/nitrate contaminants.

Thus, Logan has major deficiencies in regards to the present invention which relates to brines and what is needed to produce brines that are conducive to supporting microbial populations. First, Logan does not treat brines; Logan treats drinking water, a totally different environment. Drinking water is not a brine by definition, regardless of presence of small amount of metal salts including monovalent and divalent metal cations, as brines are toxic to humans. Second, in Logan, the divalent to monovalent ratio is naturally occurring in the ground water source and is not man made. Third,

divalent ions are not added to the water to produce the desired divalent to monovalent ratio as the ratio is naturally occurring. Fourth, the ratio is not maintained during perchlorate digestion. Fifth, the Logan water has a salinity much less than 1 (0.006% based on the milligrams of NaCl).

To the overwhelming deficiencies in Logan, the Examiner combines Logan with Okeke et al (Okeke). Okeke does indeed teach monovalent alkali metal brines, but fails to disclose, teach or even suggest adding a divalent salt to the brines to form a stabilized growth media for microbes capable of degrading per chlorates and/or nitrates. Thus, the combination of Logan and Okeke cannot render the present invention obvious for a host of reasons.

First, the combination would produce a brackish water – a mixture of fresh water and a brine, with no control over the water's salinity. Moreover, if one were to mix the AGW with the Okeke brines, one would not obtain a solution including the proper divalent to monovalent ratio as the only divalent cations would come from the AGW aquifer water, while the brines would wholly swamp the ratio by diluting the divalent metal cation concentration will below that required in the instant invention.

Second, there is no teaching in either reference to add divalent metal cations to the water to achieve and to maintain a desired divalent to monovalent metal ion molar ratio. The combination simply fails to teach or even suggest to an ordinary artisan that such a ratio is needed to produce a stable medium for microbes capable of degrading per-chlorates and/or nitrates. It is only through the teaching of the present invention that an ordinary artisan is given information sufficient to determine that a given divalent to monovalent metal ion molar ratio is needed to produce a stable growth medium for microbes capable of degrading per-chlorates and/or nitrates. Logan includes no teaching to the ratio and neither does Okeke.

Because the combination is silent on adding divalent metal cations to achieve a given molar ratio of monovalent metal to divalent metal cations, which produces a stable medium for microbes capable of degrading per chlorates and/or nitrates and maintaining the ratio during bioremediation, the combination cannot render the present claims obvious. Applicants, therefore, respectfully request withdrawal of this rejection.

11. **Claims 27-30** stand rejected under 35 U.S.C. §103 (a) as obvious over combined teachings of Logan (U.S. Patent 6,214,607 B1) in view of Okeke et al (2002. Reduction of perchlorate and nitrate by salt tolerant bacteria. ENVIRONMENTAL POLLUTION, Volume 118, Pages 357-363.,

Applicants' IDS filed 08/13/2010, Item 11) as applied to Claims 14-26 above and further in view of Venkatesh et al (U.S. Patent 6,066,257).

The Examiner states and contends as follows:

Claims 27-30 are additionally drawn to a method to degrade at least one contaminant present in a contaminated brine stream having "a salinity of  $\geq$  about 3%" when said stream is:

- fed to a bioreactor containing a mixed bacterial culture, wherein;
  - said bacterial culture degrades under anoxic/anaerobic conditions said at least one pollutant in said brine stream;
  - the column resin in said reactor is exhausted, or is not able to remove the contaminant, the flow of contaminant-containing brine is stopped to said reactor, or fed to a second reactor, while the resin in the first column is recharged with the addition of a divalent cation solution to maintain the divalent to monovalent cation molar ratio at  $\geq 0.05$ .

Regarding Claims 27-30, teachings from Logan and Okeke et al., have been discussed *supra*. Logan however, is silent regarding switching the reactors or regeneration of column resin.

Venkatesh et al., teach that the units employed in Venkatesh et al's method steps have a regeneration zone, wherein the low concentration brine is used as regenerant. Then the waste stream comprising perchlorate containing brine is treated in a bioreactor and the resulting stream is recycled (Abstract). Thus, Venkatesh et al' method remedies the discrepancy of column/resin regeneration step in Logan's method.

It would have been *prima facie* obvious to a person of ordinary skill in the art and one having ordinary skill in the art at the time of the claimed invention would have been motivated to modify/combine the teachings from Logan with those of Okeke et al., and Venkatesh et al., to obtain a method wherein at least one contaminant present in a contaminated brine stream having "a salinity of  $\geq$  about 3%" is decontaminated/reduced with a bacterial co-culture with which said brine stream is inoculated and further a method wherein the column/resin incapacitated of removing the perchlorate contaminant from the brine would be regenerated; because Okeke et al., teach a contaminant brine stream having "a salinity of  $\geq$  about 3%", 100 mg/L perchlorate in final concentration and said stream further comprises a co-culture comprising *Citrobacter* sp. and perlace, wherein said co-culture reduces the contaminant/perchlorate in said stream, and Venkatesh et al., teach regeneration of columns/resins and re-feeding the perchlorate-contaminated brine to said regenerated columns/resins. The recited prior art may be silent regarding the exact technique as are instantly claimed, however, since similar steps and same components and principle are described in the prior art references, application of those parameters and steps is deemed merely a matter of judicious design selection of a result-effective parameter which is well within the purview of the skilled artisan especially with the relevant art from the cited references before him/her as a guide (See, e.g., M.P.E.P. §2144.06) and is therefore obvious under 35 U.S.C. §103(a).

From the teachings of the cited references, it is apparent that one of ordinary skill in the art would have had a reasonable expectation of success in producing the claimed invention. Therefore, the invention as a whole was *prima facie* obvious to one of ordinary skill in the art at the time the invention was made, as evidenced by the references, especially in the absence of evidence to the contrary.

Applicants reassert their comments concerning the combination Logan and Okeke here. The inclusion of Venkatesh et al. (Venkatesh) to the combination only adds regeneration to the method. The problem is, as stated *supra*, that the combination fails to disclose, teach, even suggest or lead an ordinary artisan to add a divalent metal ion to a brine comprising a monovalent alkali metal to produce a stable medium for microbes capable of degrading per chlorates and/or nitrates and maintaining the ratio during bioremediation.

Because the combination is silent on adding divalent metal cations to achieve a given molar ratio of monovalent metal to divalent metal cations, which produces a stable medium for microbes capable of degrading per chlorates and/or nitrates and maintaining the ratio during bioremediation, the combination cannot render the present claims obvious. Applicants, therefore, respectfully request withdrawal of this rejection.

Having responded to the rejection, Applicants believe that this application is in condition for allowance.

If it would be of assistance in resolving any issues in this application, the Examiner is kindly invited to contact applicant's attorney Robert W. Strozier at 713.977.7000

**The Commissioner is authorized to charge or credit Deposit Account 501518 for any additional fees or overpayments.**

Date: 17 January 2011

Respectfully submitted,

/Robert W. Strozier/

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